NTP Research Concept: Exposure Characterization and Reproductive Health of Men Working with Bisphenol A in the United States

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Background and Rationale:
Bisphenol A (BPA) is used in the production of polycarbonated plastics and epoxy resins. There is an extensive literature on endocrine activity of BPA in various animal studies.[1] Several studies also report BPA effects on sperm in mice[2,3] and rats[3,4,5,6], decrease in free testosterone in mice[7] and one report showing BPA effects in the erectile tissue in rabbits[8]. In 2000, a consensus report prioritized chemicals tested by the National Toxicology Program (NTP) Reproductive Assessment by Continuous Breeding protocol for implementation into occupational studies of male reproductive health. BPA was prioritized with a Medium (toxicity) High (number of workers) priority.[9] Due to a lack of human studies, controversy has occurred for the past several years on the potential risk of BPA to human health[10,11]. A NTP-CERHR report expressed minimal concern for health in the occupational setting[12]; however, that report proposed critical data needs as follows: 1) human exposure assessment to clarify bisphenol A exposures and internal dosimetry in occupational-exposed individuals and 2) human studies relating adult exposure to reproduction and development, including effects on hormone levels.

Adding to the controversy, recent studies reported decreases in testosterone in men with environmental BPA exposure,[13, 14] and studies on sexual dysfunction of male workers in China[15,17] have generated a new concern for occupational male reproductive health of US workers. The first study reports that Chinese BPA-exposed workers had consistently higher risks of self-reported male sexual dysfunction in all domains of male sexual function tested by the International Index of Erectile Function (IIEF) and the Brief Male Sexual Function Inventory (BMSFI) than unexposed workers. An editorial commentary on this paper calls for more studies of sexual function of occupationally exposed men.[16] A second report from China was just published showing an association of decreased self-reported sexual function with increased urinary BPA levels.[17] One limitation of the Chinese studies is the subjective nature of questionnaire data. While the IIEF and BMSFI are a validated questionnaires used around the world including China, the addition of objective measures of erectile, sexual, and reproductive function would help to clarify the association of occupational BPA exposure and possible decrements in male reproductive and sexual function.

Study Goals:
Animal data suggests that BPA may have adverse effects on the reproductive health of men, including hormone levels, sperm quality, and penile erections. Recent studies of men in China with occupational exposures to BPA reported increased sexual dysfunction. A holistic analysis of the reproductive health of men exposed to BPA in the workplace is essential to clarify the risk of BPA exposure. This study must utilize

objective laboratory measures to assess both body burden of BPA and effects on reproductive health. Goals of the study are:

1) Determine BPA usage in industry, e.g., which industries and jobs use BPA, which tasks are associated with higher exposure.
2) Assess exposure to BPA among workers in these industries through aerosol, surface wipe, and urinary sample collection. Compare total BPA in urine between exposed and unexposed workers. Compare total BPA in urine, air, and surfaces between job tasks. Determine the correlation between air and urine BPA results.
3) Develop air and wipe sampling methods for BPA using LC-MS (liquid chromatography mass spectrometry) and LC-UV (liquid chromatography with UV detection).
4) Assess the reproductive health of men exposed to BPA in the workplace. This assessment will include a fecundity (fertility) assessment by evaluating male reproductive hormones and semen quality. Sexual function will be assessed by the International Index of Erectile Function (IIEF) questionnaire and the objective erectile measurements utilizing the Rigiscan® Plus Penis Rigidity Assessment System and biothesiometry of vibrotactile sensitivity of the penis.
5) Determine if there is a relationship between BPA exposure and reproductive health, e.g., determine if urinary BPA levels are correlated with semen quality parameters, rigiscan, vibrotactile, endocrine outcomes.

Proposed Approach:

Study Population

Sites involved in the manufacture of BPA or use of BPA (such as: polycarbonated plastics, epoxy resins, thermal paper, foundries, and other industries which are primary users of BPA) will be recruited for this study. An attempt will be made to find sites with high, medium, and low relative BPA exposures based on judgment from walk-through surveys. Additionally, an attempt will be made to include as many different industries and processes as possible. Workers from the same sites with a minimal potential for occupational exposure to BPA (office workers) will serve as the concurrent comparison group. Men will be recruited as participants from job titles with high, medium, and low BPA exposures based on judgment from walk-through surveys. Seventy-five exposed men will be recruited as well as 75 unexposed men.

Identifying and Selecting Study Sites

Industries and their industry associations, trade groups, and unions with probable worker exposure will be identified. These organizations will be sent a letter informing them of the study. They will be asked to identify facilities and types of operations with possible exposures. Industry associations or trade groups will be asked if they will share environmental sampling they have conducted.

Companies will be selected and ranked to attempt a good mix of exposures and processes. In the phone calls, we will collect information about number of employees in different types of tasks, request MSDS sheets, determine other agents that are used

with BPA, employee PPE, and gender mix. We will determine if any other agents are used concurrently at job sites that may affect reproductive health and take this fact into account in soliciting companies.

Among companies contacted we will determine which are best suited for the research, e.g., high and variable exposures, variety of processes and industries, other exposures. It is expected that approximately eight facilities will agree to a walkthrough and five of these sites will be suitable for the reproductive health study (in-depth survey). During these walkthroughs approximately 10 people will be asked to provide pre and post shift urine samples and exposure samples (air and wipe) will be collected.

Data Collection
A brief overview of the study, including the rationale and goals, will be presented to each shift of workers. A study team member will meet with each man in private to describe the study and to answer questions. Those deciding to participate will sign informed consent and be enrolled into the study. Workers will be reimbursed for their time and inconvenience. All study procedures have been effectively implemented in previous occupational health studies by this research team.

Exposure sample collections
Air and Wipe Sampling: Air and wipe sampling will be conducted during the walk-through surveys to determine feasibility during the in-depth surveys in later years

Urine: Total (free plus conjugated) urinary concentrations of BPA will be determined by collecting urine spot samples. Urine samples will be collected from all exposed and unexposed participants in the study. Timing of urine collection may change to best relate to exposure depending on specifics of job tasks and new information on variability among non-occupationally exposed individuals.

Reproductive health assessments and biological collections:
Each man will be asked to provide a blood sample, urine samples, and a semen sample.

Blood: An appointment for morning venipuncture will be arranged. Blood specimens will be used to examine endocrine function.

Each enrolled man will complete a work and health questionnaire which includes the International Index of Erectile Function Questionnaire (IIEF).

The Rigiscan® Plus will be used to assess erectile function during the normal sleep patterns of the participants.[18,19] It is a computerized monitor worn on the leg, with two loops encircling the penis, one on the base the other on the tip, used to study the penis during sleep. Men have penile erections during their sleep which provide useful physiologic information on erectile capacity.[20]

Endocrine status measurements: Male reproductive hormone levels will be measured as outcomes, and to examine endocrine function in relationship to other study outcomes. The hormones to be analyzed from the blood sample will include follicle stimulating hormone (FSH), luteinizing hormone (LH), free and total testosterone, and inhibin B. Changes in reproductive hormones may provide information on BPA endocrine action changes that might be noted in fecundity or sexual function.

Semen quality: Semen volume will be measured. Sperm motility will be assessed using computer assisted semen analysis system (CASA). Sperm concentration will be measured using the IDENT™ stain. Four microscope slides will be prepared for sperm morphology assessment. An aliquot of the whole semen will be diluted in TNE buffer and frozen for the sperm chromatin stability assay (SCSA®) analysis. Sperm viability will be determined by hypoosmotic swelling (HOS assay). Sperm morphometry will be conducted by CASA. A CLIA licensed contract laboratory will assess sperm morphology on the prepared slides. Both the traditional[21] and strict[22] morphology classifications will be performed on each specimen.

Significance and Expected Outcome:

This study will evaluate industrial populations with a range of exposures, especially high exposures, and measure environmental and biological markers of exposure determining the level of BPA exposure in US workers. After significant exposure above the known general population background is documented; this study will assess the complete reproductive health of male workers, including the reproductive endocrine health, sperm production and function, accessory sex gland function, and sexual function.

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